



Sofosbuvir in combination with peginterferon alfa-2a and ribavirin for non-cirrhotic, treatment-naive patients with genotypes 1, 2, and 3 hepatitis C infection: a randomised, double-blind, phase 2 trial

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Summary

Background Protease inhibitors have improved treatment of infection with hepatitis C virus (HCV), but dosing, a low barrier to resistance, drug interactions, and side-effects restrict their use. We assessed the safety and efficacy of sofosbuvir, a uridine nucleotide analogue, in treatment-naive patients with genotype 1–3 HCV infection.

Methods In this two-cohort, phase 2 trial, we recruited treatment-naive patients with HCV genotypes 1–3 from 22 centres in the USA. All patients were recruited between Aug 16, 2010, and Dec 13, 2010, and were eligible for inclusion if they were aged 18–70 years, had an HCV RNA concentration of 50 000 IU/mL or greater, and had no cirrhosis. We randomly allocated all eligible patients with HCV genotype 1 (cohort A) to receive sofosbuvir 200 mg, sofosbuvir 400 mg, or placebo (2:2:1) for 12 weeks in combination with peginterferon (180 µg per week) and ribavirin (1000–1200 mg daily), after which they continued peginterferon and ribavirin for an additional 12 weeks or 36 weeks (depending on viral response). Randomisation was done by use of a computer-generated randomisation sequence and patients and investigators were masked to treatment allocation until week 12. Patients with genotypes 2 or 3 (cohort B) received open-label sofosbuvir 400 mg plus peginterferon and ribavirin for 12 weeks. Our primary outcomes were safety and tolerability. Secondary efficacy analyses were by intention to treat and endpoints included sustained virological response, defined as undetectable HCV RNA at post-treatment weeks 12 and 24. This study is registered with ClinicalTrials.gov, number NCT01188772.

Findings In cohort A, 122 patients were assigned 200 mg sofosbuvir (48 patients), 400 mg sofosbuvir (48), or placebo (26). We enrolled 25 patients into cohort B. The most common adverse events—fatigue, headache, nausea, and chills—were consistent with those associated with peginterferon and ribavirin. Eight patients discontinued treatment due to adverse events, two (4%) receiving sofosbuvir 200 mg, three (6%) receiving sofosbuvir 400 mg, and three (12%) receiving placebo. In cohort A, HCV RNA was undetectable at post-treatment week 12 in 43 (90%; 95% CI 77–97) of 48 patients in the 200 mg sofosbuvir group; 43 (91%; 80–98) of 47 patients in the 400 mg sofosbuvir group, and 15 (58%; 37–77) of 26 patients in the placebo group. In cohort B, 23 (92%) of 25 patients had undetectable HCV RNA at post-treatment week 12.

Interpretation Our findings lend support to the further assessment, in phase 2 and 3 trials, of sofosbuvir 400 mg plus peginterferon and ribavirin for 12 weeks in treatment-naive patients with HCV genotype-1.

Funding Gilead Sciences.

Introduction

The 2011 approval of the NS3/4A protease inhibitors telaprevir and boceprevir for the treatment of patients with hepatitis C virus (HCV) genotype-1 has substantially improved rates of sustained virological response (SVR) compared with use of peginterferon alfa-2a (peginterferon) and ribavirin alone. In registration studies, the protease inhibitors in combination with peginterferon and ribavirin gave SVR rates between 66% and 75% in treatment-naive patients with chronic HCV genotype-1 infection.^{1,2} However, several features of regimens based on these protease inhibitors—eg, three-times-a-day

dosing, high pill burdens, complicated dosing strategies, low barrier to resistance, and many drug interactions and side-effects—restrict their use.^{3–6} Moreover, safety and effectiveness of these drugs have not been established in patients with chronic viral genotypes 2 and 3.

Sofosbuvir (formerly known as GS-7977; Gilead Sciences, Foster City, CA, USA) is a nucleotide analogue that is a potent and selective inhibitor of NS5B-directed HCV RNA replication in vitro.⁷ Sofosbuvir is a prodrug of 2'-deoxy-2'-fluoro-2'-C-methyluridine monophosphate that is converted within hepatocytes to its active uridine triphosphate form, causing chain termination during

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replication of the viral genome.⁸ In vitro, the active triphosphate inhibits recombinant NS5B polymerases from HCV genotypes 1–4 with similar half maximum inhibitory concentration values for each genotype, indicating broad activity across HCV genotypes.⁹ Also, compared with non-nucleoside inhibitors, nucleoside and nucleotide inhibitors in general have a higher resistance barrier.^{10,11} Findings from phase 1 and 2 studies suggest that sofosbuvir is well tolerated and has potent antiviral effects.^{12,13}

In this phase 2 trial, we aimed to assess the safety and tolerability of two different doses of sofosbuvir (200 mg and 400 mg) in combination with peginterferon and ribavirin in patients with HCV genotype-1, and also assessed sofosbuvir 400 mg with peginterferon and ribavirin in a separate, open-label cohort of patients with HCV genotypes 2 and 3.

Methods

Study design and participants

For this two-cohort study, we recruited previously untreated patients (aged 18–70 years) with chronic HCV genotypes 1–3 infection from 22 sites in the USA. We enrolled patients between Aug 16, 2010, and Dec 13, 2010, with the last follow-up visit on May 11, 2012. We enrolled patients with HCV genotype-1 into a randomised cohort (cohort A), patients with genotypes 2 or 3 were enrolled into a separate non-randomised, open-label cohort (cohort B).

To be eligible for enrolment, patients with HCV genotypes 1–3 had to have an HCV RNA concentration of 50 000 IU/mL or greater. HCV genotypes (and subtypes) were assessed at an independent laboratory (Cenetron Central Laboratories Ltd; Austin, TX, USA) with the VERSANT HCV Genotype 2.0 Assay (Siemens; Erlangen, Germany). Patients had a liver biopsy within 36 months before enrolment; those with cirrhosis were excluded from the trial. Inclusion criteria also included the following haematological and biochemical laboratory variables: a neutrophil count of $1.5 \times 10^9/L$ (or $\geq 1.25 \times 10^9/L$ for black patients), a haemoglobin concentration of 11 g/dL or higher in women or 12 g/dL or higher in men, a platelet count of greater than $90 \times 10^9/L$, total bilirubin within two times the upper limit of normal (21 $\mu\text{mol/L}$), and an albumin concentration of 30 g/L or lower. Patients with hepatitis B virus or HIV, psychiatric illness, pulmonary or cardiac disease, seizure disorder, or other serious comorbid disorders were excluded.

Before enrolment, the protocol and the informed consent were reviewed and approved by the institutional review board at each participating centre. The study was done in accordance with Good Clinical Practice guidelines and the Declaration of Helsinki. All patients provided written informed consent.

Randomisation and masking

We randomly assigned patients (2:2:1) with HCV-1 infection (cohort A) to receive sofosbuvir 200 mg, sofosbuvir

400 mg, or matching placebo, together with peginterferon and ribavirin for 12 weeks. We used a computer-generated randomisation sequence (generated by individuals at Pharstat Inc [Raleigh North Carolina, USA], who provided statistical support during the study but not at final analysis); allocation to treatment was done by use of an interactive online response system. Randomisation was stratified by IL28B rs12979860 single nucleotide polymorphism genotype (CC or non-CC) and baseline HCV RNA ($<800\,000$ IU/mL or $\geq 800\,000$ IU/mL). All patients concurrently received open-label peginterferon and ribavirin. Investigators and patients were masked to HCV RNA results and treatment allocation until week 12. All patients who achieved extended rapid virological response (eRVR) were unmasked before their visit at week 24 to ensure that any patients allocated to placebo were scheduled to continue treatment until week 48 unless stopping criteria were met. Cohort B was non-randomised, and treatment was open label.

Procedures

Patients in both cohorts received peginterferon 180 μg per week subcutaneously; ribavirin was dosed according to weight (ie, patients <75 kg received 1000 mg and those ≥ 75 kg received 1200 mg; ribavirin was given in two daily doses—400 mg in the morning and 600 mg in the evening for patients receiving 1000 mg a day, or 600 mg in the morning and 600 mg in the evening for patients receiving 1200 mg a day). We measured HCV RNA at each visit using the COBAS AmpliPrep/COBAS Taqman HCV test (Roche; Indianapolis, IN, USA) with a limit of detection of 15 IU/mL. All laboratory testing was done by Cenetron Laboratories (Austin, TX, USA).

Population sequencing of the HCV NS5B encoding region of the viral polymerase was done at DDL Diagnostic Laboratory (Rijswijk, Netherlands) on all pre-treatment viral samples. We did resistance monitoring in all patients who received sofosbuvir and had virological breakthrough or relapse (breakthrough defined as HCV RNA ≥ 15 IU/mL on treatment after having previously had HCV RNA <15 IU/mL on treatment, confirmed with two consecutive values or last available measurement; relapse defined as HCV RNA ≥ 15 IU/mL during the post-treatment period having achieved HCV RNA <15 IU/mL at end of treatment, confirmed with two consecutive values or last available measurement).

Use of erythropoietin-stimulating agents or granulocyte colony-stimulating factor was permitted to manage anaemia at the treating clinician's discretion only after the 12 weeks of sofosbuvir or placebo dosing.

Patients in the sofosbuvir groups who had an eRVR, defined as HCV RNA concentration below the 15 IU/mL limit of detection from weeks 4–12 received 12 additional weeks of peginterferon and ribavirin; patients receiving sofosbuvir who did not achieve an eRVR and all patients receiving placebo had an additional 36 weeks of treatment with peginterferon and ribavirin. We discontinued

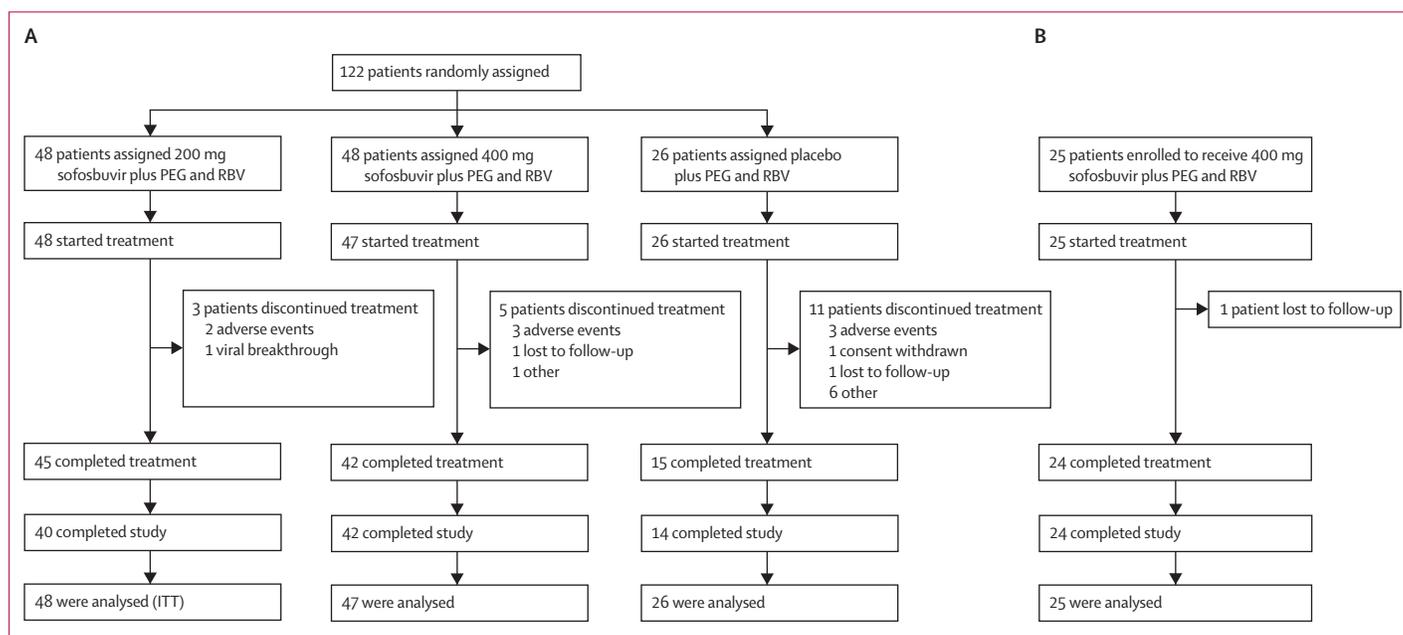


Figure 1: Trial profile

Cohort A, patients with hepatitis C genotype-1 (A). Cohort B, patients with hepatitis C genotypes 2 and 3 (B). ITT=intention to treat. PEG=peginterferon. RBV=ribavirin.

patients' treatment if they had a reduction in HCV RNA of 2 log₁₀ or less at week 12. If a patient's HCV RNA decreased by more than 2 log₁₀ by week 12, but was not below the limit of detection, we continued their treatment until week 24. If HCV RNA was still detectable at week 24, we discontinued treatment with peginterferon and ribavirin. In parallel, we enrolled a separate cohort of patients with genotype 2 or 3 in which all patients received open-label sofosbuvir 400 mg with peginterferon and ribavirin for 12 weeks, with no response-guided treatment.

Statistical analyses

We calculated the sample size for cohort A (patients with HCV genotype-1 who were randomly allocated to treatment) on the basis of the efficacy endpoint of eRVR. Assuming that 15% of patients in the control group and at least 55% of patients in the sofosbuvir groups had an eRVR, a sample size of 75 patients was sufficient to achieve 95% power to establish superiority with a two-sided significance level of 5%.

We included all randomly allocated patients who received at least one dose of study drug in all safety and efficacy analyses. The primary objective was safety and tolerability. For safety, we assessed the occurrence of self-reported adverse events (using the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events; version 1.0), clinical laboratory tests, physical examinations, vital signs measurements, and electrocardiogram readings at various timepoints during the study (appendix). The study was not designed to statistically assess efficacy. Efficacy endpoints included RVR (defined as HCV RNA <15 IU/mL at week 4 of

treatment), eRVR, response at end of treatment, and sustained virological response, defined as an HCV RNA concentration of 15 IU/mL or lower at 12 weeks after completion of treatment (SVR12) and at 24 weeks after treatment (SVR24). We tested the differences between the number of patients having an SVR in the treatment and placebo groups using the Cochran-Mantel-Haenszel test.

A data safety monitoring Board reviewed the ongoing safety of the study and met after the last patient was randomly allocated and also when the last patient completed their visit at week 12. The board consisted of three independent outside board members (including the chairman) and an independent, unmasked statistician. We used SAS (version 9.2) for all statistical analyses.

This study is registered with ClinicalTrials.gov, number NCT01188772.

Role of the funding source

The sponsor of the study contributed to recruitment of patients, trial management, data collection, statistical analyses, and the writing and review of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit the manuscript for publication.

Results

We randomly allocated 122 patients to treatment in cohort A and enrolled 25 patients into cohort B (figure 1; see appendix for details of those not enrolled). Baseline characteristics were much the same across treatment groups in cohort A: the mean age of patients was about 50 years and most patients were men, white,

See Online for appendix

	Cohort A		Cohort B	
	Sofosbuvir 200 mg plus PEG and RBV (n=48)	Sofosbuvir 400 mg plus PEG and RBV (n=47)	Placebo plus PEG and RBV (n=26)	Sofosbuvir 400 mg plus PEG and RBV (n=25)
Mean age in years (SD)	48.4 (11.5)	51.4 (9.4)	48.6 (9.4)	47.2 (11.1)
Men	33 (69%)	21 (45%)	19 (73%)	16 (64%)
Race				
White	39 (81%)	37 (78%)	21 (80%)	20 (80%)
Black	6 (13%)	7 (15%)	5 (19%)	4 (16%)
Other	3 (6%)	3 (6%)	0	1 (4%)
Ethnic origin				
Hispanic	5 (10%)	6 (13%)	1 (4%)	8 (32%)
Non-Hispanic	43 (90%)	41 (87%)	24 (92%)	17 (68%)
Not reported	0	0	1 (4%)	0
Mean body-mass index in kg/m ² (SD)	26.6 (3.4)	26.8 (4.5)	28.6 (4.1)	28.6 (4.8)
Mean log ₁₀ hepatitis C RNA in IU/mL (SD)	6.5 (0.6)	6.4 (0.8)	6.5 (0.8)	6.1 (0.8)
HCV genotype				
1a	37 (77%)	35 (74%)	20 (77%)	..
1b	11 (23%)	12 (26%)	6 (23%)	..
2	15 (60%)
3	10 (40%)
IL28b				
CC	21 (44%)	18 (38%)	11 (42%)	7 (28%)
CT	24 (50%)	19 (40%)	11 (42%)	17 (68%)
TT	3 (6%)	10 (21%)	4 (15%)	1 (4%)
Fibrosis stage				
No or minimal fibrosis	12 (25%)	7 (15%)	3 (12%)	7 (28%)
Portal fibrosis	35 (73%)	38 (81%)	21 (81%)	18 (72%)
Bridging fibrosis	1 (2%)	2 (4%)	2 (8%)	0

Data are n (%), unless otherwise stated. PEG=peginterferon. RBV=ribavirin.

Table 1: Baseline characteristics

non-Hispanic, had the IL28B CC or CT genotype, and had portal fibrosis (table 1). Most patients had genotype-1a HCV with baseline HCV RNA concentrations of about 6.5 log₁₀ IU/mL (table 1). Baseline characteristics (other than HCV genotype) were much the same between individuals in cohort A and cohort B (table 1).

The most common adverse events during sofosbuvir dosing (up to week 12) were fatigue, headache, nausea, chills, pain, and insomnia (table 2). Most adverse events were mild or moderate in severity. Adverse events seen in the sofosbuvir groups were generally consistent with events seen during treatment with peginterferon and ribavirin. Fatigue, rash, fever, and diarrhoea were more common in both sofosbuvir groups in cohort A than in the placebo group in cohort A and in cohort B, although we detected no dose-response effect (ie, events were not more common in patients receiving 400 mg of sofosbuvir compared with in those receiving 200 mg). In cohort A, a greater proportion of patients in the placebo group had headache than did in either sofosbuvir group (table 2).

Changes in laboratory values were also generally consistent with those historically associated with peginterferon and ribavirin treatment (table 3). We recorded no substantial differences between the treatment groups in both cohorts with regards to decreases in haemoglobin concentrations, or numbers of lymphocytes, leucocytes, neutrophils, or platelets (data not shown), although proportions of patients with low leucocyte and lymphocyte counts were slightly higher in patients receiving sofosbuvir than in those receiving placebo.

Three patients (one in the sofosbuvir 200 mg group and two in the sofosbuvir 400 mg group) had grade 3 or greater increases in aspartate aminotransferase concentrations, with associated increases in alanine aminotransferase concentrations to grade 2 within 4 weeks of beginning study treatment. The concentrations of these aminotransferases remained increased in these patients after the end of sofosbuvir dosing, and returned to normal (between 10 IU/L and 44 IU/L) after they had finished peginterferon and ribavirin treatment.

Eight patients in cohort A discontinued treatment because of an adverse event, six within the first 12 weeks of treatment (three in the placebo group and three in the 400 mg sofosbuvir group). Reasons for these early discontinuations are as follows: in the 200 mg sofosbuvir group, one patient had neutropenia, and one had folliculitis; in the 400 mg sofosbuvir group, one patient had an aphthous ulcer, one had a myocardial infarction, and one had worsening depression and suicidal ideation; in the placebo group, one patient had insomnia, body aches, forgetfulness, fatigue, mood swings, confusion, and arthralgia, one had shortness of breath and chest pains, and one had irritability and headache. Three other treatment-emergent serious adverse events occurred in three patients after the conclusion of sofosbuvir dosing: retinal vein occlusion (in the sofosbuvir 200 mg group on day 97 of the study), lymphangitis (in the sofosbuvir 400 mg group), and chest pain and ECG ST segment elevation (in the placebo group). None of the 25 patients in cohort B discontinued treatment because of an adverse event (figure 1).

Patients in all sofosbuvir groups had rapid and substantial reductions in HCV RNA concentrations within the first few weeks of treatment (figure 2). In cohort A, nearly all patients in the sofosbuvir groups had an RVR, whereas only a few in the placebo group had an RVR (table 4). Of the two patients receiving sofosbuvir who did not have an RVR, one (in the 400 mg group) discontinued treatment after 2 weeks because of an adverse event (suicidal ideation) and was lost to follow-up, the other (in the 200 mg group), had serum HCV RNA of 49 IU/mL at week 4—as per protocol, this patient was treated for an additional 36 weeks with peginterferon and ribavirin and achieved SVR24. In cohort B, all but one patient achieved an RVR (table 4). The patient who did not achieve RVR was lost to follow-up after taking at least one dose of study drug.

In cohort A, compared with the placebo group, SVR12 and SVR24 were more common in the 200 mg sofosbuvir group (differences of 30%, 95% CI 12–49, $p=0.001$, and 28%, 9–46, $p=0.0017$, respectively) and in the 400 mg sofosbuvir group (differences of 32%, 13–51; $p=0.0005$, and 30%, 11–49, $p=0.0006$, respectively; table 4). The reason that fewer patients achieved SVR24 than achieved SVR12 in both sofosbuvir groups in cohort A was not because of viral relapse: two of the three patients, both in the 200 mg group, were lost to follow-up after achieving SVR12; the other patient, who was in the 400 mg group, was erroneously scheduled for their post-treatment week-24 visit 3 days before the beginning of the window for that visit—that patient's test result were therefore recorded as SVR12 and further follow-up was not possible.

Of the 25 patients in cohort B, most achieved both SVR12 and SVR24 (table 4). Of the two patients who did not achieve SVR, one was lost to follow-up after the baseline visit and the other had undetectable HCV RNA at post-treatment weeks 12 and 24, but we did not classify these results as SVR12 and SVR24 because the assays were done in a commercial laboratory that used an HCV RNA concentration of 43 IU/mL as the cutoff for detectability (the protocol definition of undetectable was <15 IU/mL). Four patients, all in cohort A, who discontinued treatment early achieved SVR12 and SVR24; two from the 200 mg sofosbuvir group after 19–23 weeks of treatment, and two from the 400 mg sofosbuvir group after 11–14 weeks of treatment.

No patient in any sofosbuvir group had virological breakthrough during sofosbuvir treatment. Three patients with HCV genotype-1 in the 200 mg sofosbuvir group had a breakthrough after completion of sofosbuvir dosing while receiving peginterferon and ribavirin. One patient in the control group had viral breakthrough at week 20 of peginterferon and ribavirin treatment. None of the patients receiving sofosbuvir had viral non-response to treatment; nine patients in the placebo group did not respond at all to treatment.

Two patients receiving sofosbuvir (both in cohort A; one in the 200 mg group and one in the 400 mg group) had viral relapse within 4 weeks of completing the full course of treatment (table 4). Two patients in the 400 mg group had treatment failure after early discontinuation of treatment (one received 2 weeks and the other received 6 weeks of treatment).

Previous in-vitro studies identified NS5B Ser282Thr as the primary sofosbuvir-resistance mutation.¹⁰ Changes in the NS5B polymerase in clinical isolates from six of seven patients who either relapsed after completion of treatment (two), discontinued early (two), or who had virological breakthrough during peginterferon and ribavirin treatment (three) were successfully obtained by population sequencing. HCV RNA could not be sequenced in one patient because of low viral load at the time of relapse (177 IU/mL). No Ser282Thr mutation was detected at the virological failure timepoint by population sequencing,

	Cohort A			Cohort B
	Sofosbuvir 200 mg plus PEG and RBV (n=48)	Sofosbuvir 400 mg plus PEG and RBV (n=47)	Placebo plus PEG and RBV (n=26)	Sofosbuvir 400 mg plus PEG and RBV (n=25)
Fatigue	32 (67%)	32 (68%)	14 (54%)	9 (36%)
Headache	17 (35%)	18 (38%)	15 (58%)	11 (44%)
Nausea	15 (31%)	21 (45%)	9 (35%)	12 (48%)
Chills	18 (38%)	19 (40%)	10 (38%)	9 (36%)
Pain	14 (29%)	12 (26%)	8 (31%)	4 (16%)
Insomnia	9 (19%)	14 (30%)	8 (31%)	4 (16%)
Rash	13 (27%)	12 (26%)	4 (15%)	3 (12%)
Neutropenia	8 (17%)	14 (30%)	5 (19%)	6 (24%)
Anaemia	11 (23%)	8 (17%)	5 (19%)	3 (12%)
Muscle pain	5 (10%)	11 (23%)	6 (23%)	5 (20%)
Fever	12 (25%)	10 (21%)	2 (8%)	4 (16%)
Arthralgia	5 (10%)	5 (11%)	5 (19%)	8 (32%)
Diarrhoea	7 (15%)	11 (23%)	2 (8%)	1 (4%)

Data are n (%). Adverse events that occurred in more than 20% of patients in each group are listed. PEG=peginterferon. RBV=ribavirin. *Data show adverse events collected until week 12 of sofosbuvir treatment.

Table 2: Treatment-emergent adverse events*

	Cohort A			Cohort B
	Sofosbuvir 200 mg plus PEG and RBV (n=48)	Sofosbuvir 400 mg plus PEG and RBV (n=47)	Placebo plus PEG and RBV (n=26)	Sofosbuvir 400 mg plus PEG and RBV (n=25)
Haemoglobin				
Grade 3 (70 to <90 g/L or ≥ 45 g/L decrease from baseline)	14 (29%)	8 (17%)	4 (15%)	4 (17%)
Leucocytes				
Grade 3 (1.0×10^9 /L to 1.5×10^9 /L)	3 (6%)	4 (9%)	1 (4%)	0
Lymphocytes				
Grade 3 (0.35×10^9 /L to 0.5×10^9 /L)	2 (4%)	2 (4%)	1 (4%)	1 (4%)
Grade 4 ($<0.035 \times 10^9$ /L)	2 (4%)	0	0	0
Neutrophils				
Grade 3 (0.5×10^9 /L to 0.75×10^9 /L)	10 (21%)	15 (32%)	5 (19%)	6 (25%)
Grade 4 ($<0.5 \times 10^9$ /L)	2 (4%)	0	0	0
Aspartate aminotransferase (IU/L)				
Grade 3 ($>5 \times$ ULN to $10 \times$ ULN)	1 (2%)	1 (2%)	0	0
Grade 4 ($>10 \times$ ULN)	0	1 (2%)	0	0
Total bilirubin				
Grade 3 ($>2.5 \times$ ULN to $5 \times$ ULN)	1 (2%)	0	0	1 (4%)

PEG=peginterferon. RBV=ribavirin. ULN=upper limit of normal. *Data show grade 3 or 4 haematology and hepatological laboratory parameters until week 12 of sofosbuvir treatment.

Table 3: Haematological and hepatological laboratory parameters*

and no mutation at the other conserved residues was consistently detected in patients with virological failure. Drug susceptibility data were successfully generated from five patients, but were not available from the other two patients (one because of low viral load and the other because of poor replication). We detected no change in half maximum effective concentration values for sofosbuvir from any of these five patients at the time of virological failure.

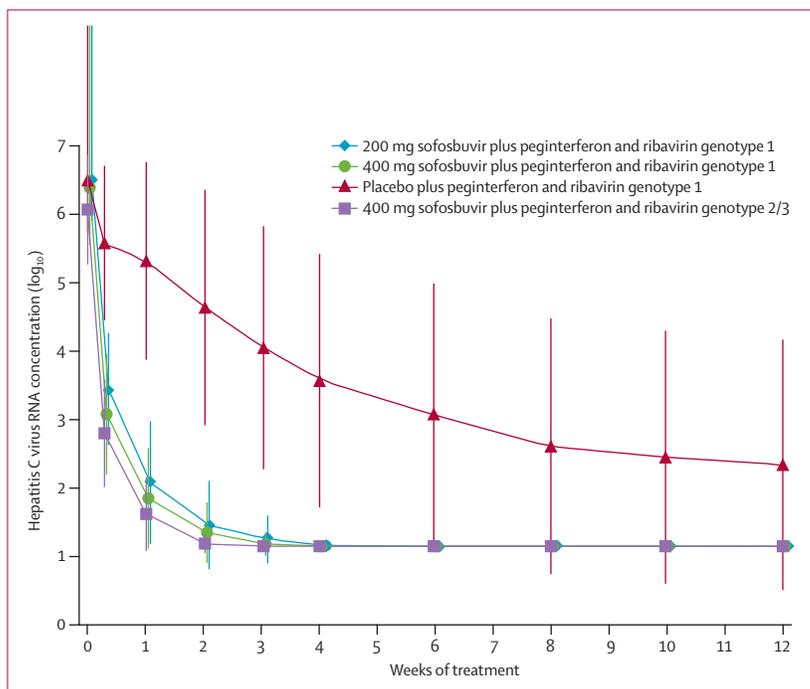


Figure 2: Mean concentrations of hepatitis C virus RNA during the 12 weeks of sofosbuvir treatment, by treatment group

	Cohort A			Cohort B
	Sofosbuvir 200 mg plus PEG and RBV (n=48)	Sofosbuvir 400 mg plus PEG and RBV (n=47)	Placebo plus PEG and RBV (n=26)	Sofosbuvir 400 mg plus PEG and RBV (n=25)
RVR	47 (98%; 89–100)	46 (98%; 89–100)	5 (19%; 7–39)	24 (96%; 80–100)
End of week 12	48 (100%; 93–100)	43 (91%; 80–98)	16 (62%; 41–80)	24 (100%; 86–100)
End of treatment	45 (94%; 83–99)	47 (100%; 93–100)	16 (62%; 41–80)	24 (100%; 86–100)
SVR12	43 (90%; 77–97)	43 (91%; 80–98)	15 (58%; 40–77)	23 (92%; 74–99)
SVR24*	41 (85%; 72–94)	42 (89%; 77–96)	15 (58%; 40–77)	23 (92%; 74–99)
Virological failure				
Virological rebound or breakthrough	3 (6%)	0	2 (8%)	0
Post-treatment relapse	1 (2%)	1 (2%)	0	0

Data are n (%; 95% CI) or n (%). PEG=peginterferon. RBV=ribavirin. RVR=rapid virological response (undetectable hepatitis C RNA at week 4). SVR12=sustained virological response at week 12 after treatment. SVR24=sustained virological response at week 24 after treatment. *Two patients from the 200 mg group and one from the 400 mg group who achieved SVR12 had missing values for post-treatment week 24—we did a sensitivity analysis in which these three patients were excluded from the denominator of the SVR24 estimate: on the basis of this analysis, 41 of 46 patients (89%, 95% CI 76–96%) in the 200 mg group and 42 of 46 patients (91%, 95% CI: 79–98%) for the 400 mg group achieved SVR24.

Table 4: Virological response during treatment and follow-up (intention-to-treat population)

Discussion

In this phase 2 trial, patients receiving sofosbuvir plus peginterferon and ribavirin had adverse events that were similar in both type and severity to those seen in patients receiving placebo plus peginterferon and ribavirin, with an adverse event profile broadly consistent with that seen elsewhere during treatment

with peginterferon and ribavirin.^{14–16} The most common events—fatigue, headache, nausea, and chills—are well known side-effects of interferon. We detected no additional or new adverse events attributable to sofosbuvir. Fatigue, rash, fever, and diarrhoea were more commonly seen in patients with HCV genotype-1 receiving sofosbuvir than in patients receiving placebo, but we recorded no dose-response effect (ie, no difference in the occurrence of these adverse events between the 200 mg and 400 mg sofosbuvir groups). Moreover, all of these events except fever were less common in patients with HCV genotype-1 receiving sofosbuvir than they were in patients with HCV genotype-2 or genotype-3 receiving sofosbuvir (table 2). The number of patients with decreased leucocyte and lymphocyte counts was slightly higher in patients with genotype-1 receiving sofosbuvir, but the number of events was too small for any definite conclusions to be drawn. None of these safety signals were seen in a cumulative analysis of safety data from more than 500 patients who received sofosbuvir in phase 2 trials.¹⁷ The most pronounced differences in the occurrence of adverse events in that study were associated with the presence or absence of peginterferon. Suppression of neutrophils and haemoglobin was substantially greater in patients receiving interferon than in those receiving only sofosbuvir or sofosbuvir and ribavirin.¹⁷

Sofosbuvir plus peginterferon and ribavirin provided rapid and durable suppression of HCV RNA in large proportions of treatment-naive patients with HCV genotypes 1–3. After 4 weeks of treatment, 96–98% of patients receiving sofosbuvir had undetectable HCV RNA. As a result of this high rate of early response, all but one patient treated with sofosbuvir qualified for the shorter course of treatment (24 weeks vs 48 weeks). In view of this result, response-guided treatment based on early response is unlikely to be useful with sofosbuvir-based regimens.

During the sofosbuvir phase of treatment, response in patients with HCV genotype-1 receiving sofosbuvir was much the same, irrespective of dose (200 mg or 400 mg; figure 2). However, differences emerged during the peginterferon and ribavirin phase of dosing. Three patients in the 200 mg group had viral breakthrough while on peginterferon and ribavirin, two within 4 weeks and one within 8 weeks of the end of sofosbuvir treatment. By contrast with these findings, no patients in the 400 mg group had virological breakthrough, suggesting that the 400 mg dose might provide more effective viral suppression. All three of the patients in the 200 mg group who had virological breakthrough had a baseline HCV RNA of greater than 800 000 IU/mL and the CT IL28B allele.

In the intention-to-treat analysis of patients with HCV genotype-1, 90% of patients in the 200 mg group and 91% of patients in the 400 mg achieved SVR12, with 85% in the 200 mg group and 89% in the 400 mg group achieving SVR24. These rates of response compare

Panel: Research in context**Systematic Review**

We consulted three reviews on new and emerging pharmacotherapies for chronic hepatitis C virus (HCV) infection,^{3,5,18} and searched PubMed for studies written in English using the search term “HCV treatment”. We did our last search on Jan 18, 2003. We identified many clinical trials that were still in progress, most of which assessing direct-acting antiviral agents, including protease inhibitors, NS5A inhibitors, NS5B nucleoside analogues, NS5B non-nucleoside inhibitors, and various combinations of these agents with and without peginterferon and ribavirin. Additionally, we reviewed treatment guidelines for hepatitis C.^{19,20}

Interpretation

For treatment-naïve patients with genotype-1 HCV, the standard-of-care consists of one of two protease inhibitors—telaprevir or boceprevir—plus peginterferon and ribavirin; the duration of treatment is between 24 weeks and 48 weeks, depending on patient’s response to treatment. With these regimens, 66–75% of treatment-naïve patients achieved rates of sustained virological response.¹² Although such rates of response are a substantial improvement compared with the rate achieved with peginterferon and ribavirin alone, the former standard of care, the protease inhibitor regimens leave several medical needs unmet. Our data lend support to the further assessment of a 12 week regimen of sofosbuvir 400 mg in combination with peginterferon and ribavirin for the treatment of patients with HCV genotype-1, and in interferon-free regimens for patients with HCV genotypes 2 and 3.

favourably with those seen with current standard-of-care treatment (panel). The slightly lower rates of SVR24 than SVR12 in these group was not due to known viral relapse, but to factors unrelated to the antiviral efficacy of the regimen, suggesting that concordance between SVR12 and SVR24 would closely approach 100% with more thorough follow-up. The 30% difference in outcomes between sofosbuvir-containing groups and the peginterferon and ribavirin control seems similar to the difference between the experimental and control groups in phase 3 trials of the protease inhibitors telaprevir (75% vs 44%) and boceprevir (67–68% vs 40%).¹² However, the 58% SVR rate for the control group in this study was higher than the historical average of 40–50%,²¹ which might be partly due to the exclusion of patients with cirrhosis.

In patients with HCV genotypes 2 and 3 in cohort B, 23 (92%) of 25 patients had both SVR12 and SVR24. In this group, no viral breakthrough occurred during any phase of treatment and no patient relapsed after the end of treatment; the two patients who are not counted among those achieving SVR did not have treatment failure. These data seem to align with results from the ELECTRON study,²² in which all patients with genotype 2

or 3 treated with sofosbuvir 400 mg with or without peginterferon and ribavirin for 12 weeks achieved SVR24. These findings also seem to substantiate the suggestion that 12 weeks of treatment is sufficient in patients with HCV genotypes 2 and 3.¹³

An important limitation of this study was that it excluded patients with cirrhosis, a factor that has been associated with reduced response to treatment. In a small study (N=16),²³ the initial rate of HCV RNA decay in patients with hepatic impairment seemed to be slower than that in patients with HCV infection but without cirrhosis. However, a median decrease of about 3.6 log₁₀ IU/mL was seen in these patients with cirrhosis over 7 days, indicating that sofosbuvir has strong antiviral activity. Ultimately, response rates in patients with cirrhosis will depend on HCV genotype, the drug or drugs that sofosbuvir is combined with, and the duration of treatment.

Our data suggest further testing of the 400 mg once daily dose of sofosbuvir in interferon-containing and interferon-free regimens for a total duration of 12 weeks across HCV genotypes, and that these regimens should be tested in a broader population of patients, including those with cirrhosis. In the ongoing phase 3 study (NCT01641640), we are assessing sofosbuvir in combination with peginterferon and ribavirin for 12 weeks in treatment-naïve patients with HCV genotype-1.

Contributors

EL, JPL, TH, KVK, FFP, NHA, DEB, EDJ, BF, DRN, DTD, IMJ, DJ, JMD, MR-T, KRR, MSS, NHB, RHH, HM, EA, WTS, MMB, and AM contributed to the writing and review of the report. JPL, TH, KVK, DRN, IMJ, DJ, GAA, MR-T, and KRR contributed to recruitment of patients. JPL, TH, KVK, BF, IMJ, MR-T, and KRR were investigators in this study. EL, JPL, TH, KVK, AMS, NHA, DEB, ED, JMD, KRR, MSS, HM, MM, RHH, and RH contributed to the data collection. EL, KVK, FFP, AMS, NHA, ED, DRN, GAA, KRR, MSS, HM, ML, RH, EA, WTS, RHH, and AM contributed to the data interpretation. EL, DEB, DRN, DJ, MSS, MM, RH, WTS, MMB, RHH, and AM contributed to the study design. EA and MMB contributed to medical oversight.

Conflicts of interest

EL has received research support and grants from Abbott, Achillion, Anadys, Biolog, Boehringer Ingelheim, Bristol-Myers Squibb, Gilead, GlaxoSmithKline, GlobeImmune, Idenix, Idera, Inhibitex, Intercept, Janssen, Medarex, Medtronic, Merck, Novartis, Pharmasset, Roche, Schering-Plough, Santaris, Scynexis, Vertex, ViroChem, and ZymoGenetics; was on the speakers’ bureau for Gilead, Merck, and Vertex; and has served on advisory boards for Abbott, Achillion, Anadys, Biolog, Biotica, GlobeImmune, Inhibitex, Merck, Novartis, Pharmasset, Tibotec, Theravance, and Vertex. TH has received research grants from Bristol-Myers Squibb, Abbott Pharmaceuticals, Gilead Sciences, Janssen Pharmaceuticals, Icaria Pharmaceuticals, Takeda Pharmaceuticals, Mochida Pharmaceuticals, Sundise Pharmaceuticals, Roche Pharmaceuticals, Eisai Pharmaceuticals, Vertex Pharmaceuticals; and has received honoraria as a speaker for Gilead. KVK has received research support and grants from Abbot, Beckman, Boehringer Ingelheim, Bristol-Myers Squibb, Conatus, Gilead, Icaria, Intercept, Janssen, Merck, Mochida, and Vertex; has served as a consultant to Exjade; has received honoraria (speaking and advisory board) from Merck and Novartis; and has served on advisory boards for Abbott, Gilead, and Vertex. FFP has received research grants from Achillion, Anadys, Boehringer Ingelheim, Bristol-Myers Squibb, Genentech, Gilead, Human Genome Science, Idenix, Inhibitex, Janssen, Merck, Novartis, Pharmasset, Salix, and Vertex; has served as a consultant or adviser to Achillion, Anadys, Boehringer Ingelheim, Bristol-Myers Squibb, Genentech, Gilead, Idenix, Inhibitex, Janssen, Kadmon, Merck, Novartis, Salix, and Vertex; and was on the

speakers' bureau for Genentech, Gilead, Kadmon, Merck, Salix, and Vertex. AMS has received research grants from Gilead and Pharmasset. NHA has received research support from Abbott, Echosens, Gilead, GlaxoSmithKline, Novartis, Pharmasset, Quest, Schering-Plough/Merck, and Vertex; has received honoraria (consultant and adviser) from Boehringer Ingelheim, Echosens, Gilead, GlaxoSmithKline, Ligand, Medgenics, Novartis, Springback, and Vertex. DEB has received honoraria for serving on speakers' bureau from Gilead. ED has served on advisory boards and has received honoraria from Gilead. BF has received research support from Gilead. DRN has received research support and has served as a consultant and adviser to Gilead. DTD has received research support, served as a consultant, and was on the speakers' bureau for Achillion, Boehringer Ingelheim, Bristol-Myers Squibb, Genentech, Gilead, Idenix, and Merck. IMJ has received research support and grants from Abbott, Achillion, Anadys, Boehringer Ingelheim, Bristol-Myers Squibb, Genentech, Gilead, Janssen, Novartis, Pfizer, Schering-Plough/Merck, and Vertex; was on the speakers' bureau for Bristol-Myers Squibb, Genentech, Gilead, Schering-Plough/Merck, and Vertex; and has received consultancy fees from Abbott, Achillion, Boehringer Ingelheim, Bristol-Myers Squibb, Enanta, Genentech, Gilead, GlaxoSmithKline, Idenix, Janssen, Kadmon, Novartis, Presidio, Schering-Plough/Merck, and Vertex. DJ has received research grants from Abbott, Boehringer Ingelheim, Bristol-Myers Squibb, Genentech, and Janssen; and has served on advisory boards for Abbott, Astex, Biotica, Boehringer Ingelheim, Bristol-Myers Squibb, Genentech, Gilead, Inhibitex, Janssen, Merck, and Vertex. MRT has received research support and grants from Abbott, Akros, Anadys, Beckman Coulter, Boehringer Ingelheim, Bristol-Myers Squibb, Genentech, Gilead, GlaxoSmithKline, Hoffman-La Roche, Human Genome Sciences, Idenix, Idera, Inhibitex, Johnson and Johnson, Merck, Mochida, Novartis, Pfizer, Pharmasset, Santaris, Scynexis, Siemens Healthcare Diagnostics, Vertex, and Zymogenetics; and has served as a consultant for Akros, Bristol-Myers Squibb, Genentech, Hoffman-La Roche, Inhibitex, Janssen, Merck, Pharmasset, Santaris, and Vertex. KRR has received research support and grants from Anadys, Bristol-Myers Squibb, Genentech, Gilead, Ikaria, Janssen, Merck, and Vertex; and has served on advisory boards for Bristol-Myers Squibb, Genentech, Gilead, Janssen, Merck, Novartis, and Vertex. MSS has received research grants from Abbott, BIPI, Bristol-Myers Squibb, Gilead, Janssen, Merck, Roche, and Vertex; and has served as a consultant to Abbott, BIPI, Bristol-Myers Squibb, Gilead, Janssen, Merck, Novartis, Pfizer, Roche, and Vertex. NHB has received research support from Bristol-Myers Squibb, Genentech, Gilead, Johnson and Johnson, Merck, Pharmasset, Vertex, and Zymogene; has served as a consultant to Abbott; and was on the advisory boards and speakers' bureau for Gilead and Vertex. RHH, HM, ML, MM, RH, EA, WTS, and MMB are employees and stockholders of Gilead Sciences. AM has received research grants from Abbott, Achillion, Bristol-Myers Squibb, Gilead, GlaxoSmithKline, Medtronic, Merck, Pfizer, Scynexis, and Vertex; and has served as a consultant to Achillion, GlaxoSmithKline, Merck, Scynexis, and Vertex. JPL, GAA, and JMD declare that they have no conflicts of interest.

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